

02/16/2009

Appln. No. 10/579,032

Reply dated February 9, 2009

Reply to Office Action dated October 8, 2008

Amendments to the Claims:

This listing of the claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims:

1-3. (Canceled).

4. (Currently Amended) The method according to claim \pm 31, wherein the resulting microparticles have an average particle diameter of 0.01. μm to 150 μm .

5. (Currently Amended) The method according to claim \pm 31, wherein the resulting microparticle is a drug carrier.

6. (Currently Amended) The method according to claim \pm 31, wherein the resulting microparticle is a sustained-release drug carrier.

7. (Currently Amended) The method according to claim \pm 31, wherein the dilute solution before the crosslinking reaction contains a drug, and the drug is held in microparticles obtained after the crosslinking reaction.

8. (Original) The method according to claim 7, wherein the crosslinking reaction does not cause drug denaturation even in the presence of the drug.

9-10. (Canceled).

11. (Withdrawn) The method according to claim 1, wherein the crosslinking reaction is a reaction in which crosslinkages are formed by reaction between hydrazide group and an activated carboxylic acid ester.

12-19. (Canceled).

20. (Withdrawn) The microparticle according to claim 12, wherein the crosslinkage functional group is a mercapto group, and the crosslinking reaction is a reaction in which crosslinkages are formed by disulfide formation.

21. (Canceled).

22. (Withdrawn) The microparticle according to claim 12, wherein the crosslinking reaction is a reaction in which crosslinkages are formed by reaction between a hydrazide group and an activated carboxylic acid ester.

23. (Canceled).

24. (Currently Amended) The method according to claim ~~23~~ 4, wherein the resulting microparticle is a drug carrier.

25. (Previously Presented) The method according to claim 24, wherein the resulting microparticle is a sustained-release drug carrier.

26. (Previously Presented) the method according to claim 25, wherein the dilute solution before the crosslinking reaction contains a drug, and the drug is held in the microparticles obtained after the crosslinking reaction.

27. (Previously Presented) The method according to claim 26, wherein the crosslinking reaction does not cause drug denaturation even in the presence of the drug.

28-30. (Canceled).

31. (New) A method for preparing crosslinked polysaccharide microparticles, which comprise the following steps:

a) preparing a dilute solution containing (1) a polysaccharide derivative having at least one crosslinkage functional group in a range of 0.1 to 5% (w/v) and (2) a crosslinking agent;

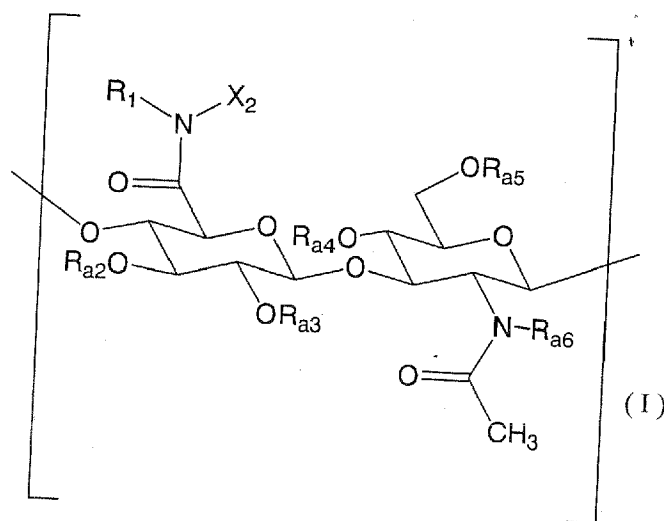
b) dispersing the solution by spraying to form microparticulate droplets; and

c) concentrating the solution contained in the droplets to facilitate a crosslinking addition reaction of the polysaccharide derivative between a mercapto group and a unsaturated C-C bond;

wherein steps b) and c) are carried in a spray drying procedure;

wherein the polysaccharide derivative is a hyaluronic acid derivative comprising at least one repeating unit represented by Formula (I);

[Formula I]



wherein X_2 represents $-Y_1-Q_1-N(-R_2)-Y_3-Q_2-SH$, $-N(-R_2)-Y_3-Q_2-SH$, $-NHCO-(CH_2)_4-CONH-NH-C(=NH)-(CH_2)_3-SH$, $-(CH_2)_2-NH-C(-NH)-(CH_2)_3-SH$, or $-(CH_2)_2-O-(CH_2)_2-O-(CH_2)_2-NH-C(=NH)-(CH_2)_3-SH$,

R_1 represents a hydrogen atom, a linear or branched C_{1-10} alkyl group, a linear or branched C_{1-10} hydroxyalkyl group, a polyalkylene oxide group, a polypeptide group or a polyester group,

R_{a2} , R_{a3} , R_{a4} , R_{a5} and R_{a6} each independently represent a hydrogen atom, a linear or branched C_{1-6} alkyl group, a linear or branched C_{1-6} alkenyl group, a linear or branched C_{1-16} alkynyl group, a linear or branched C_{1-16} alkylcarbonyl group, a linear or branched C_{1-6} alkenylcarbonyl group, a linear or branched C_{1-16} alkynylcarbonyl group or $-SO_2OH$,

Y_1 represents a single bond, $-N(-R_3)CO-$, $-N(-R_3)-$, $-CO-$ or $-CH_2CO-$,

Y_2 represents a single bond, $-CON(-R_4)-$ or $-N(-R_4)-$,

Q_1 represents a linear or branched C_{1-10} alkylene group, a linear or branched C_{1-10} hydroxyalkylene group, a polyalkylene oxide group, a polypeptide group or a polyester group,

R_2 , R_3 and R_4 each independently represent a hydrogen atom, a linear or branched C_{1-10} alkyl group, a linear or

branched C₁₋₁₀ hydroxyalkyl group, a polyalkylene oxide group, a polypeptide group or a polyester group,

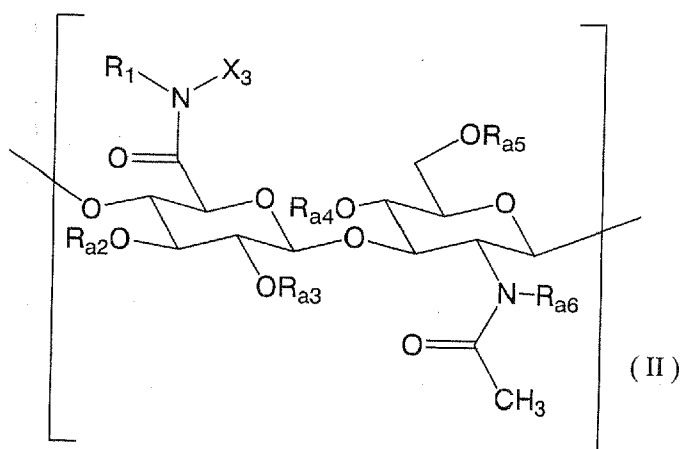
Y₃ represents a single bond, -CO-, -CO₂-, -CH₂-CH(OH)- or -CONH- and

Q₂ represents a linear or branched C₁₋₁₀ alkylene group, a linear or branched C₁₋₁₀ hydroxyalkylene group, a polyalkylene oxide group, a polypeptide group or a polyester group,

and the crosslinking agent is a compound having two or more unsaturated C-C bond-containing groups; or

the polysaccharide derivative is a hyaluronic acid derivative comprising at least one repeating unit represent by Formula (II):

[Formula 2]



wherein X₃ represents -Y₁, Q₁-Y₂-N(-R₂)-Y₃-Q₄ or -n(-R₂)-Y₃-Q₄,

R_1 represents a hydrogen atom, a linear or branched C_{1-10} alkyl group, a linear or branched C_{1-10} hydroxyalkyl group, a polyalkylene oxide group, a polypeptide group or a polyester group,

R_{a2} , R_{a3} , R_{a4} , R_{a5} and R_{a6} each independently represent a hydrogen atom, a linear or branched C_{1-6} alkyl group, a linear or branched C_{1-6} alkenyl group, a linear or branched C_{1-16} alkynyl group, a linear or branched C_{1-16} alkylcarbonyl group, a linear or branched C_{1-6} alkenylcarbonyl group, a linear or branched C_{1-16} alkynylcarbonyl group or $-SO_2OH$,

Y_1 represents a single bond, $-N(-R_3)CO-$, $-N(-R_3)-$, $-CO-$ or $-CH_2CO-$,

Y_2 represents a single bond, $-CON(-R_4)-$ or $-N(-R_4)-$,

Y_3 represents a single bond, $-CO-$ or $-CH_2CO-$,

Q_1 represents a linear or branched C_{1-10} alkylene group, a linear or branched C_{1-10} hydroxyalkylene group, a polyalkylene oxide group, a polypeptide group or a polyester group,

R_2 , R_3 and R_4 each independently represent a hydrogen atom, a linear or branched C_{1-10} alkyl group, a linear or branched C_{1-10} hydroxyalkyl group, a polyalkylene oxide group, a polypeptide group or a polyester group,

Q_4 represents a linear or branched C_{2-10} alkenyl group, a linear or branched C_{2-10} alkynyl group,

and the crosslinking agent is a compound having two or more mercapto groups.

32. (New) The method according to claim 5, wherein the crosslinked polysaccharide microparticles are injectable.

33. (New) The method according to claim 5, wherein the drug is a protein.

34. (New) The method according to claim 6, wherein the sustained release period of the carrier is 24 hours or more.

35. (New) The method according to claim 6, wherein the sustained release period of the carrier is 5 days or more.

36. (New) The method according to claim 6, wherein the drug is released upon enzymatic digestion.